

# PATENT COOPERATION TREATY



## PCT

### INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

(Chapter II of the Patent Cooperation Treaty)

(PCT Article 36 and Rule 70)

REC'D 02 AUG 2005
WIPO PCT

Applicant's or agent's file reference 10451.204-WO		<b>FOR FURTHER ACTION</b>		See Form PCT/PEAA16
International application No. PCT/DK2004/000231		International filing date (day/month/year) 02.04.2004	Priority date (day/month/year) 04.04.2003	
International Patent Classification (IPC) or national classification and IPC C12N1/00, C12P19/14, C12P7/06, C13K1/06				
Applicant NOVOZYMES AS et al.				
<p>1. This report is the international preliminary examination report, established by this International Preliminary Examining Authority under Article 35 and transmitted to the applicant according to Article 36.</p> <p>2. This REPORT consists of a total of 4 sheets, including this cover sheet.</p> <p>3. This report is also accompanied by ANNEXES, comprising:</p> <p>a. <input checked="" type="checkbox"/> sent to the applicant and to the International Bureau) a total of 2 sheets, as follows:</p> <p><input checked="" type="checkbox"/> sheets of the description, claims and/or drawings which have been amended and are the basis of this report and/or sheets containing rectifications authorized by this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions).</p> <p><input type="checkbox"/> sheets which supersede earlier sheets, but which this Authority considers contain an amendment that goes beyond the disclosure in the international application as filed, as indicated in item 4 of Box No. I and the Supplemental Box.</p> <p>b. <input type="checkbox"/> (sent to the International Bureau only) a total of (indicate type and number of electronic carrier(s)) , containing a sequence listing and/or tables related thereto, in computer readable form only, as indicated in the Supplemental Box Relating to Sequence Listing (see Section 802 of the Administrative Instructions).</p>				
<p>4. This report contains indications relating to the following items:</p> <p><input checked="" type="checkbox"/> Box No. I Basis of the opinion</p> <p><input type="checkbox"/> Box No. II Priority</p> <p><input type="checkbox"/> Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability</p> <p><input type="checkbox"/> Box No. IV Lack of unity of invention</p> <p><input checked="" type="checkbox"/> Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement</p> <p><input type="checkbox"/> Box No. VI Certain documents cited</p> <p><input type="checkbox"/> Box No. VII Certain defects in the international application</p> <p><input type="checkbox"/> Box No. VIII Certain observations on the international application</p>				
Date of submission of the demand 28.01.2005		Date of completion of this report 02.08.2005		
Name and mailing address of the International preliminary examining authority:  European Patent Office - P.B. 5818 Patentlaan 2 NL-2280 HV Rijswijk - Pays Bas Tel. +31 70 340 - 2040 Tx: 31 651 epo nl Fax: +31 70 340 - 3016		Authorized Officer Lejeune, R Telephone No. +31 70 340-2347 		

# INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International application No.  
PCT/DK2004/000231

## Box No. I Basis of the report

1. With regard to the **language**, this report is based on the international application in the language in which it was filed, unless otherwise indicated under this item.
  - ☐ This report is based on translations from the original language into the following language , which is the language of a translation furnished for the purposes of:
    - ☐ international search (under Rules 12.3 and 23.1(b))
    - ☐ publication of the international application (under Rule 12.4)
    - ☐ international preliminary examination (under Rules 55.2 and/or 55.3)
2. With regard to the **elements\*** of the international application, this report is based on *(replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report)*:

### Description, Pages

1-15 as originally filed

### Claims, Numbers

1-12 received on 28.01.2005 with letter of 28.01.2005

- ☐ a sequence listing and/or any related table(s) - see Supplemental Box Relating to Sequence Listing
3. ☐ The amendments have resulted in the cancellation of:
    - ☐ the description, pages
    - ☐ the claims, Nos.
    - ☐ the drawings, sheets/figs
    - ☐ the sequence listing (*specify*):
    - ☐ any table(s) related to sequence listing (*specify*):
  4. ☐ This report has been established as if (some of) the amendments annexed to this report and listed below had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).
    - ☐ the description, pages
    - ☐ the claims, Nos.
    - ☐ the drawings, sheets/figs
    - ☐ the sequence listing (*specify*):
    - ☐ any table(s) related to sequence listing (*specify*):

\* If item 4 applies, some or all of these sheets may be marked "superseded."

**INTERNATIONAL PRELIMINARY REPORT  
ON PATENTABILITY**

International application No.  
PCT/DK2004/000231

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**Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement**

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**1. Statement**

Novelty (N)	Yes: Claims	1-12
	No: Claims	
Inventive step (IS)	Yes: Claims	
	No: Claims	1-12
Industrial applicability (IA)	Yes: Claims	1-19
	No: Claims	

**2. Citations and explanations (Rule 70.7):**

**see separate sheet**

**Re Item V.**

The following documents are referred to in this communication:

D1 : WO 97/42301 A (GIST BROCADES BV ;LAROYE MARIE PAULE (FR);  
SOUPE JEROME (FR)) 13 November 1997 (1997-11-13)

**Novelty (Art 33(2) PCT)**

D1 discloses a method for the production of wort comprising the provision of a mash comprising starch, where the mash is heated in a number of steps from 50 to 76 degrees C in the presence of alpha-amylase, beta-glucanase and xylanase (see examples). The preliquefaction, gelatinisation and liquefaction take place successively during the period where the mash is heated.

The subject matter of claim 1 (and for essentially the same reasons claims 2-12) is new because D1 does not disclose a gelatinization step by jet cooking or a step for the recovering of the ethanol.

**Inventive step (Art 33(3) PCT)**

The subject matter of claims 1-12 does not involve an inventive step because the differences between the present application and D1 (gelatinization step by jet cooking or a step for the recovering of the ethanol) are standard procedures in the field of hydrolysis of starch compounds and production of ethanol.

## Claims

1. A method of producing ethanol, said method comprising the steps of:
  - a. providing a mash comprising a starch containing material and water;
  - b. preliquefying the mash of step (a) in the presence of a beta-glucanase;
  - c. gelatinizing the mash of step (b) by jet cooking;
  - d. liquefying the mash of step (c) in the presence of an alpha-amylase, a beta-glucanase and a xylanase; and
  - e. saccharifying and fermenting the mash of step (d) to produce ethanol.
  - f. recovering the ethanol.
2. The method of claim 1, further comprising a pre-saccharification step which is performed after the liquefaction step (d) and before step (e).
3. The method of any of claims 1 or 2, wherein the xylanase is derived from a strain of *Aspergillus* sp., preferably from a strain of *A. Aculeatus*.
4. The method of any of claims 1-3, wherein the beta-glucanase is derived from a strain of *Bacillus* sp., preferably from a strain of *B. amyloliquefaciens*.
5. The method of any of claims 1-4, wherein also an endo-glucanase is present in the liquefaction step (d), said endo-glucanase preferably derived from a strain of *Trichoderma* sp., preferably from a strain of *T.reesei*.
6. The method of any of claims 1-5, wherein the starch containing material is obtained from cereals and/or tubers.
7. The method of any of claims 1-6, wherein the starch containing material is selected from the groups consisting of maize, wheat, barley, rye, millet, sorghum, and milo.
8. The method of any of claims 1-7, wherein the starch containing material is selected from the groups consisting of potato, sweet potato, cassava, tapioca, sago, banana, sugar beet and sugar cane.
9. The method of any of claims 1-8, wherein the fermentation in step (e) is performed using a micro-organism, such as bacteria and fungi (including yeasts), e.g. *Zymomonas* species and *Sacharomyces* species. such as e.g. *Saccharomyces cerevisiae*.
10. The method of any of claims 1-9, wherein the fermentation is carried out in the presence of phytase and/or protease.
11. The method of any of claims 1-10, wherein preliquefaction in step (b) is performed at

a temperature of 45 to 70°C, more preferably to 53 to 66°C, most preferably to 55 to 60°C, such as 58°C for a period of 5 to 60 minutes, and more preferably 10 to 30 minutes, such as around 15 minutes.

12. The method of any of claims 1-11, wherein the liquefaction in step (d) is performed at 60-95°C, preferably 80-90°C for 10-120 min, more preferably at 83-85 °C for 15-80 min.